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Behavioural and clinical responses of turkeys stunned in a V-shaped, carbon dioxide tunnel

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Abstract

*Stunning with carbon dioxide gas (CO_2) is used in turkeys (*Meleagris gallopavo var domesticus*) as an alternative to electrical water bath stunning. Investigations were carried out to assess the behavioural and clinical responses of turkeys stunned in a V-shaped CO_2 tunnel while sitting in their transport crates as part of the usual slaughter routine in a commercial abattoir. The CO_2 concentration in the tunnel rose from 27% at the first observation point (A) to 74% at point D and the transport time was 180 s. At window 1, 37.1% of the birds showed head shaking, 3.7% deep breaths and 2.9% intensive wing-flapping and at window 2 the respective figures were 2.2, 18.4 and 6.2%. All birds appeared to have lost consciousness at the point of leaving the tunnel. Prior to shackling, 15 s after leaving the tunnel, 230 animals were tested for both interphalangeal reflex and eyelid closure; 20.4% had an incomplete eyelid closure but none displayed an interphalangeal reflex. It would appear that the tunnel system we investigated stuns turkeys effectively within 180 s. However, the initial stunning phase of 40–105 s appears to cause the animals distress, which is demonstrated by head shaking, deep breaths and wing flapping.*

Keywords: *animal welfare, behaviour, CO_2 stunning, reflexes, slaughter, turkeys*

Introduction

The worldwide production of turkey meat has risen in recent years from 1,200,000 tonnes in 1970 to in excess of 5,100,000 tonnes in 2005 (Windhorst 2005). For example, in 2005, the EU produced approximately 1,890,000 tonnes and the USA 2,464,000 tonnes (Anonymous 2006a). Turkeys tend to be slaughtered in specialised slaughter plants. The most common method of stunning turkeys is via electrical waterbath stunning (Abeyesinghe *et al* 2007). In Germany, the use of carbon dioxide (CO_2) gas for stunning turkeys requires a permit from the relevant local veterinary office which supervises the slaughterhouse (Anonymous 2006b).

The main welfare benefits of CO_2 stunning compared with electrical stunning were listed recently by the World Organisation for Animal Health (Office Internationale des Epizooties [OIE 2007]):

• Gas stunning of poultry is applied while the animals remain in their transport containers; there is no need for uncrating.

• Birds are not handled and shackled while conscious.

• All the birds in a crate are exposed to the stunning gas at the same time. No bird can be avoided as can be the case with electrical waterbath stunning (Simmonds 2004).

It is important to clarify the impact of CO_2 on the birds' physiology as this underpins any understanding of turkey behaviour in the tunnel and these effects are probably similar to those seen in mammals (Raj 2006). Firstly, there is a rise in respiratory rate and depth of breathing which results in a higher uptake of CO_2 (Eisele *et al* 1976). Secondly, stimulation of receptors in the glomus aorticum and glomus caroticum occurs, activating the vasomotor centre and leading to vasoconstriction (as investigated in pigs by Cantieni 1977). Thirdly, it leads to a drop in the pH of blood and cerebrospinal fluid which causes the narcotic effect of CO_2 (as seen in dogs by Eisele *et al* 1976). At the same time, the drop in pH leads to a reduction in the force of the heart's contraction, causing central vasoconstriction and peripheral vasodilatation (as investigated in pigs by Mullenax & Dougherty 1963). In addition, birds have CO_2-sensitive, intrapulmonary chemoreceptors (Raj 2006) which can also affect the rate and volume of breathing; a rise in CO_2 concentration causes stimulation of the vagus nerve, leading to a depressed respiratory rate. As well as these physiological responses, clinical and behavioural reactions can also be observed in birds which may help an assessment of consciousness levels. Typical reactions include head shaking and gasping; both of which are associated with the experience of unpleasant sensations during CO_2 inhalation (Raj 1996). These reactions are considered to occur while animals are conscious (Schäfer 1995). The movement of the

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head into opisthotonus is associated with excitations, visible as convulsive wing flapping, which occur in a state of unconsciousness (Schäfer 1995; Raj 2006) and the greater the initial concentration of CO_2 the earlier the onset of these convulsions (Raj & Gregory 1990).

Korbel (1998) developed a protocol to evaluate the narcotic status and extent to which birds are unconscious and listed typical reflexes, physical signals and behavioural reactions, including eyelid closure and interphalangeal reflex; an absent interphalangeal reflex and completely closed eyes (Barton Gade *et al* 2001a) were interpreted as signs of deep unconsciousness.

Different tunnel systems exist for the stunning of poultry with CO_2 gas:

• CAS (controlled atmosphere stunning) tunnels with subsequent chambers including different gas concentrations (McKeegan *et al* 2007a).

• Two-tiered tunnel systems that feature increasing CO_2 concentrations (Drawer 2007).

• V-shaped tunnels with rising CO_2 concentration (von Wenzlawowicz *et al* 2000).

Von Wenzlawowicz *et al* (2000) made similar investigations in one of the first V-shaped tunnels. Here, plastic curtains were installed, not only in the tunnel but also at the tunnel entrance and exit, to create different concentrations of CO_2 and O_2. The authors made mention of the need for further investigation as they figured out elements of the stunning system which could be optimised, especially in the initial stunning phase.

In order to assess the status of the birds in the V-shaped tunnel two approaches were taken: (i) observing and describing behavioural responses of turkeys on entering and encountering the CO_2 atmosphere in an open V-shaped stunning tunnel, under commercial conditions and (ii) assessing the level of consciousness/unconsciousness after traversing the tunnel and prior to bleeding to death, by checking the interphalangeal reflex and eyelid closure under practical conditions.

Materials and methods

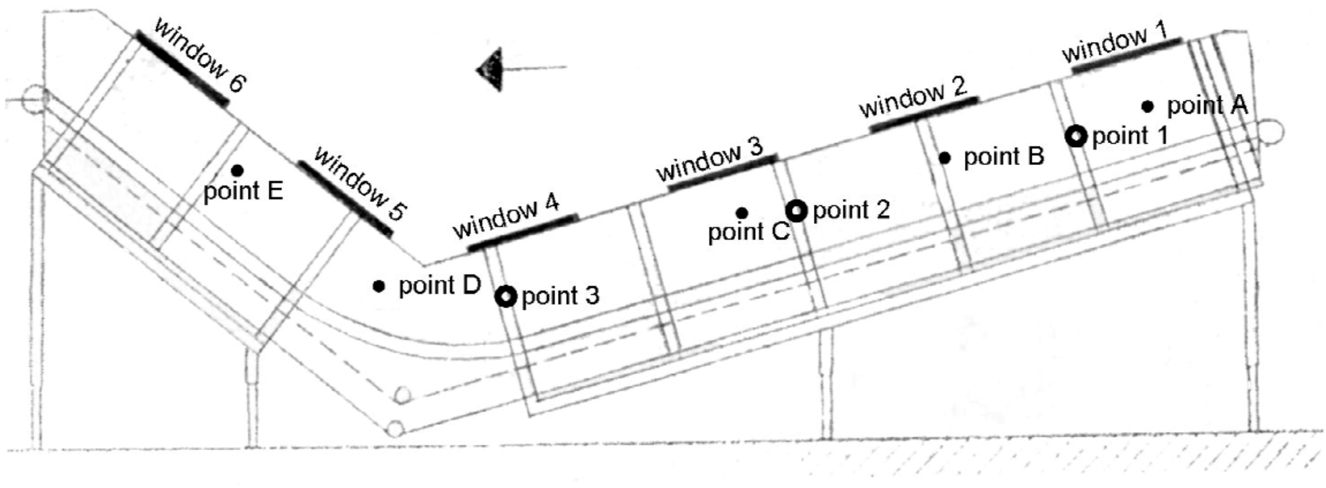
The slaughterhouse and stunning system

This study was carried out in a medium-sized, commercial poultry slaughterhouse in Brenz, in the north-east of Germany which slaughters, on average, 8,000 turkeys per day. Slaughtering begins at 0545h and ends between 1300 and 1400h depending on the workload and males tend to be slaughtered first in the morning, followed by the hens. The turkeys are transported in standardised (see below) crates to the slaughterhouse on lorries specifically designed for carrying these crates. They originated from farms ranging from 11 to 313 km from the plant. The crates (115 × 75 × 33 cm; length × width × height) are plastic with no top lid; the bottom of the upper crate serving as a lid for the crate below. Top crates are closed by transport frames and the sides and floor have perforations for ventilation. Each crate transports either eight hens (1,078 cm^2 per bird) or five cocks (1,725 cm^2 per bird). On arrival at the plant, the frames are unloaded automatically onto a mechanical conveyor system which transfers single boxes continuously into the gas tunnel. The V-shaped gas tunnel (YARA, Dülmen, Germany) is stainless steel and measures 9.1 × 1.4 × 1.0 m (length × width × height) (see Figure 1). The descending portion is 6.1 m in length while the ascending part is 3.0 m. The entrance descends at a gradient of 13° and the exit ascends at 32°. The open, top segment of the tunnel is equipped with plexiglass windows (80 × 60 cm; length × width) to facilitate observation; these can also be opened for cleaning and gaining access to the birds. There are a total of four windows in the descending part (windows 1–4) and two in the ascending part (windows 5–6). These two parts of the tunnel have two separate conveyor belts; one descending and one ascending which transport crates at the same speed. Turkeys passing through the tunnel in the crates are exposed to increasing levels of CO_2 with the highest concentration occurring at the bottom of the tunnel (D). One crate takes 180 s to pass through the tunnel. It takes 15 s for a crate to reach the centre of the first window after entering the tunnel. Between two consecutive window centres the time span is 30 s and finally a crate emerges from the tunnel 15 s after having passed window 6. The transit time from point A to D is 105 s and from D to E, 30 s. At the ascending end of the tunnel, the boxes are automatically tipped and the birds fall onto a conveyor belt which transports them to the point of shackling. Before the turkeys are shackled manually, the interphalangeal reflex and eyelid closure are tested. After bleeding to death, the eviscerated carcases are placed in a cold store with an air temperature of 2 to 3°C.

Gas monitoring

The CO_2 was injected into the tunnel at points 1–3 (Figure 1) in the descending portion. CO_2 concentration was measured regularly at points A–E (Figure 1) by the EL6010-Uras14 analyser (ABB-Advance Optima Systems, Zurich, Switzerland) using non-dispersive infrared (NDIR) absorption which selectively measures CO_2 via photometry. In parallel, oxygen (O_2) was measured at the same points using the EL6010-Magnos106 analyser (ABB-Advance Optima Systems, Zurich, Switzerland). This measuring principle is based on the specific paramagnetic behaviour of oxygen. Both gases were measured continuously for approximately four hours throughout the tunnel on five typical working days. The tube inlets, through which the gas samples were taken, were placed at points A–E (Figure 1) in the tunnel. The gas was removed by pumps at a continuous rate of 60 l min^{-1} through Teflon tubes (4 × 1 mm; inner diameter × wall thickness) from the sampling point to the analyser. A selective unit directed the gas from the different sampling points to the analyser at a fixed time schedule in order that a measurement could be performed every five seconds. Both units of the gas analyser were calibrated according to the manufacturer's manual, using nitrogen, CO_2 and ambient air as calibration gas.

Figure 1



The V-shaped CO_2 tunnel for stunning turkeys showing the positions of gas inlet points (1–3), gas measuring points (A–E) and observation windows (1–6).

The animals

A total of 1,790 turkeys (713 males, 1,077 females) of the strain BUT Big 6 were investigated. The average age of the males was 20 to 21 weeks and their weight was approximately 20 kg. The hens were slaughtered at an age of 16 weeks, at a bodyweight of approximately seven-to-nine kg. For practical reasons, it was not possible to carry out behavioural observations (through tunnel windows) and clinical examinations (prior to shackling) on the same birds. However, the birds showed a high degree of homogeneity in age and weight and they underwent the same transport and abattoir procedures.

Behavioural observations

The reactions of 960 females and 600 males (n_1) were observed directly through instantaneous scan sampling by two individuals through the four windows of the descending part of the tunnel (windows 1–4) and at the tunnel's exit, during two normal working days. Observers sat on the tunnel and looked through the glass windows and stood next to the tunnel exit where birds were tipped out. One crate was always fully visible for 16 s in a window. Every bird was observed once and the behavioural reaction noted. No bird was monitored twice in a crate. Occasionally, one or two birds in a crate were disguised by standing birds and not fully visible. Each observer monitored a single window for a timespan of 10 min, after which point they changed positions in order that both monitored an equal number of birds at each window section and at the tunnel exit. The total time spent on direct observations at each window and at the tunnel's exit was approximately 100 min, divided into 50 min of hen observations (n = 960) and 50 min of cock observations (n = 600). As the concentration of CO_2 increased, the following behavioural reactions and head and body postures were monitored at the four observation windows and at the end of the tunnel prior to tipping out:

- Wing-flapping, divided into 'slight' (only slight single flaps without a complete wing stretching) and 'intense' flapping (flaps were more frequent and powerful, but still single and not continuous, wings were stretched during flapping as much as possible in such a small space).
- Deep breaths (a long, deep breath with opened beak).
- Head shaking (a short but intensive head shake).
- A change of head position from normal to opisthotonus (the head moves into opisthotonus at the moment of monitoring).
- Convulsive wing flapping (excitations shown as long running, ceaseless flaps with a high frequency, single flaps are unable to be distinguished).
- Loss of head posture (the head is totally atonic).
- Preserved head posture (control of head posture still apparent).

Slight wing flapping at the beginning of the tunnel was interpreted as a reaction to the tilting of the crates when entering the decline of the tunnel, with birds attempting to keep their balance. Thereafter, wing flapping became more vigorous and could also be interpreted as a defensive reaction, along with head shaking, to the rising CO_2 concentration. Head shaking probably occurred as a result of the acid sensation of CO_2 on mucosal membranes (Raj 1996) and may be an indication of disorientation and/or irritation (Abeyesinghe *et al* 2007). Deep breaths, occurring as a physiological response to rising levels of CO_2, are suggested to be due more to dyspnoea than pain, but are still viewed as a welfare concern since they occur during the conscious phase (Abeyesinghe *et al* 2007). The

Table 1 Median concentrations and quartiles (25 and 75) of CO_2 and O_2 at five different points (A–E) in the tunnel.

Point	CO_2 (% by volume)	O_2 (% by volume)
A	27 (25: 28)	16 (15.8: 16.3)
B	35 (31: 37)	14.5 (14: 15)
C	67.5 (65: 68)	8 (7.6: 8.3)
D	74 (73: 75)	6.5 (6: 7)
E	71 (70: 72)	7.5 (7:8)

move into opisthotonus is an indication that unconsciousness is being reached; it follows on from convulsive wing flapping until a loss of head posture signals unconsciousness (Barton Gade *et al* 2001a).

Assessment of interphalangeal reflex and eyelid closure

During the normal slaughter routine 230 birds (n_2) (113 cocks and 117 hens) were randomly taken from the conveyor belt and tested for eyelid closure and interphalangeal reflex, as according to Korbel (1998). These two parameters are also regarded as reliable parameters for unconsciousness by other researchers (eg Sinn 1994; Lawton 1996). Both parameters were relatively quick and easy to assess during the small time period between the end of stunning and the start of shackling. For the interphalangeal reflex test, the interdigital skin of both feet was subjected to painful stimuli using surgical tweezers and assessed as either negative (no reaction) or positive (reaction). Eyelid closure was assessed in three categories: closed, half open and fully open. After this inspection, which lasted less than one minute per animal, the birds were placed back onto the conveyor belt to continue the normal slaughtering process.

Results

Gas monitoring

Table 1 shows the median gas concentrations and the 25:75% quartiles at the five measuring points (A–E) of all measurements. The CO_2 concentration rises from an average of 27% at point A to 74% at point D before declining at the end of the tunnel to 71% at point E. O_2 concentration dropped from 16% at point A to 6.5% at point D, increasing to 7.5% at point E. These gas concentrations were relatively stable and variations remained low.

Behaviour

Table 2 summarises the results of behavioural observations at windows 1–4 and the tunnel exit. The majority of turkeys were seen to be sitting in the crates as they made their way to the tunnel entrance and as they entered the stunning tunnel. Many showed signs of discomfort, such as opened-beak breathing and tongue movements (Barton Gade *et al* 2001a) which were probably a result of the stress of transport, high temperatures and noise in the slaughterhouse.

Head shaking was seen mainly in window 1 (37.1%) and the number of sightings reduced significantly in the two following windows. In total, head shaking was seen for a period of about 60 s.

Approximately half of the birds demonstrated slight wing flapping in the first window. This may have been another initial reaction to CO_2 but as it occurred in direct conjunction with the tilting of the crate, we feel it is more likely to be linked to the birds' attempts to keep their balance (Barton Gade *et al* 2001b).

Deep breaths (18.4%) and intensive wing flapping (6.2%) both peaked in the second window, appearing in window four 75 s after being seen in window 1 for the first time.

The movement towards opisthotonus was observed mainly in window 2 (12%) and here, also, we first noted a loss of head posture and convulsive wing flapping. Both of these behavioural reactions peaked in window 3, 75 s after the beginning of stunning. In window 4, 9.2% of the turkeys were yet to lose head posture but, 75 s later at the end of the tunnel, all birds were motionless in the crates.

Tests for unconsciousness after the tunnel

Figure 2 clarifies the results of the examination of eyelid closure. Nearly 80% of the birds (183 turkeys) had their eyes shut when leaving the stunning tunnel. The 20% (46 turkeys) with half-opened eyes and the one bird with opened eyes (0.4%) might have partly resulted from the handling of the birds. When moving the carcasses, the skin of the birds may have been dragged upwards, causing the eyelid to change position. No animal gave a positive reaction to the painful interphalangeal reflex. This may be an indication that the animals were either in a deep stage of unconsciousness or, perhaps, even dead when leaving the stunning tunnel.

Discussion

The time taken for turkeys to reach unconsciousness and the manner in which they do so has implications for welfare in general and the stunning process in particular. Coenen *et al* (2005) demand the painless elimination of consciousness by inducing a quick and adequate anaesthesia for the first stage of a slaughter protocol.

With increased CO_2 in the descending part of the tunnel, typical reactions were observed. Head shaking indicates discomfort and can be attributed to irritation of mucous membranes by the acidic CO_2 (Raj 1994). Also, McKeegan *et al* (2007b) concluded that CO_2 has aversive properties. Slight, as well as vigorous, wing-flapping in the course of the stunning procedure could have been a conscious reaction to this unpleasant sensation.

Also, deep breaths were observed mainly in the stunning phase, where we suggest most of the birds were still conscious. These deep breaths can be seen as the earliest indicator of respiratory distress in poultry (Raj 2006) therefore the inhalation of hypercapnic gas mixtures is likely to be an unpleasant and disconcerting experience for birds (McKeegan *et al* 2007b).

Table 2 Percentages of turkeys (n = 1,560) showing different behavioural responses to increasing levels of CO_2 at four points in the tunnel (windows 1–4) and at the tunnel exit.

Behavioural response	Window 1	Window 2	Window 3	Window 4	Tunnel exit
Head shaking	37.1	2.2	0.4	0	0
Slight wing flapping	47.6	4.7	9.2	30.4	0
Intensive wing flapping	2.9	6.2	2.1	1	0
Deep breaths	3.7	18.4	2.4	1.7	0
Head moves towards opisthotonus	0	12	5.5	3.4	0
Convulsive wing flapping	0	5.2	30.6	4.9	0
Lost head posture	0	3	10.4	not counted	not counted
Preserved head posture	not counted	not counted	not counted	9.2	0

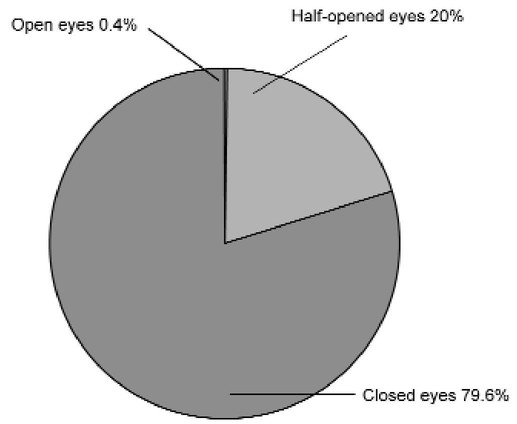
The simultaneous vigorous wing flapping observed here also underlines this discomfort and should be viewed as defensive moves, perhaps even as attempts to escape this unpleasant situation. It is our opinion, therefore, that this initial phase is detrimental to animal welfare. The majority of the turkeys appear to experience these behavioural reactions while conscious.

Taking opisthotonus as an indication of the onset of unconsciousness (Barton Gade *et al* 2001a), we observed this after only 45 s in window 2 and it was quickly followed by a loss of head posture (Barton Gade *et al* 2001a) and excitations manifest as convulsive wing flaps; a clear indication of unconsciousness (Raj 2006).

By window 4, most of the birds had suffered a loss of posture, not only from the head but also the rest of the body, and a large amount of slight wing flapping was also noted (30.4% of all birds) which may be interpreted as final movements after the convulsions. The excitations shown as convulsive wing flapping did not stop abruptly but faded out becoming slight wing flapping once more. These flaps were qualitatively different from the slight wing flaps seen at the beginning of the stunning process as it appeared that the birds were unconscious and the flaps were not seen as relevant to animal welfare. Despite this, we should note that unconsciousness does not begin simultaneously for all birds but that it is reached individually. Individual factors, such as differing bodyweight or differences in the respiratory tract, may delay or enhance the absorption of CO_2. Indeed, 9.2% of turkeys had not lost head posture in window 4. This creates a further welfare concern since these turkeys may still be capable of experiencing distress and discomfort and (presumably even) pain, 105 s after entering the stunning tunnel. Additionally, it is suggested that the short-lived experience of seeing other birds convulse and of being struck by bodies and flapping wings might also negatively impact on bird welfare (Raj 2006).

Therefore, it was important to also assess the stunning result: at the end of the tunnel all the turkeys had reached a deep state of unconsciousness and no bird regained consciousness during bleeding. It appeared that many of the birds were already dead at the clinical inspection prior to shackling. This assumption is supported by the results of the interphalangeal reflex test and the eyelid closure test.

Figure 2



Percentage of eyelid closure observed in turkeys (n_2 = 230) after CO_2 stunning and prior to shackling.

Conclusions and animal welfare implications

• The stunning of turkeys in this V-shaped tunnel, via increasing concentrations of CO_2, is effective and allows easy shackling and bleeding of unconscious birds.

• It would appear that increasing CO_2 concentration from 27–74%, over a period of 180 s, reliably stuns male and female turkeys for slaughter.

• At the entrance to the tunnel, when the birds enter the CO_2 atmosphere, typical signs of distress and discomfort were observed for a period of between 45 and 105 s.

• The heterogeneous onset of reaching unconsciousness in this type of stunning tunnel has to be mentioned and critically assessed in terms of animal welfare.

• CO_2 stunning has considerable potential for the effective stunning of turkeys. However, the initial stunning phase appears suboptimal and should be improved. In addition, an earlier and more uniform timepoint for the onset of unconsciousness would be desirable.

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